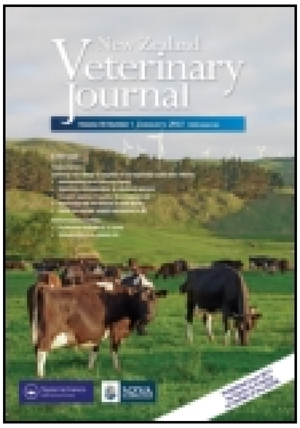

New Zealand Veterinary Journal
Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/tnzv20

Rotavirus and coronavirus associated diarrohoea in calves

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Published online: 23 Feb 2011.

To cite this article: P.J.K. Durham , B.C. Farquharson & B.J. Stevenson (1979): Rotavirus and coronavirus associated diarrohoea in calves, New Zealand Veterinary Journal, 27:12, 266-272

To link to this article: http://dx.doi.org/10.1080/00480169.1979.34669

Rotavirus and coronavirus associated diarrohoea in calves

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N.Z.vet.J.27: 266 & 271-2

ABSTRACT

A virological survey was carried out on 150 calf faeces from 30 central North Island properties, utilising electron miscroscopic examination of ultracentrifuged faecal pellets. Rotaviruses were detected on three farms, and coronavirus-like particles on two farms.

In the same area, three other properties with past histories of rotavirus infection were also studied. Weekly examination of faeces from 10 calves from each property over a period of 5 weeks demonstrated that rotavirus excretion commenced at the same time as the onset of outbreaks of diarrhoea. On two of these rotavirus-positive farms, coronaviruses were also detected. At one farm, this coincided with rotavirus excretion and diarrhoea, and was associated with a more severe clinical disease but, on the other property, the coronavirus was unassociated with clinical disease. A similar study, carried out on a further property with a past history of coronavirus infection, showed that coronavirus excretion commenced simultaneously with the onset of an outbreak of diarrhoea. The virus was, however, detected in small numbers in 1 of 10 calves only.

The results of the survey, coupled with previously gathered data, indicate that both viruses may be widespread in the cattle population. On some properties, rotavirus had a clear association with diarrhoea, but the relationship of coronavirus to diarrhoea was less well-established.

INTRODUCTION

Neonatal diarrhoea of calves is a problem which has troubled farmers for many years. Though *Escherichia coli* has frequently been blamed in the past, recent reports[14][18] have cast some doubt on the extent to which this organism can be incriminated. It is now realised that the induction of clinical disease with *E. coli* is dependent not only on the strain involved, but also on the numbers and distribution of the organism within the intestine[1][10].

The incrimination in recent years of a number of newly recognised viruses as causative agents of diarrhoea in young animals has led to a much better appreciation of the complex aetiology of calf diarrhoea. In particular, it has led to the realisation that a variety of infective agents may be involved, sometimes in combination; and that these may interact with a number of host and environmental factors. These latter may predispose to, or prevent, the development of infection or of clinical disease.

Because overseas reports have incriminated rotaviruses[10] and coronaviruses[12] as causative agents of diarrhoea in calves, increased attention has been devoted in New Zealand towards the electron-microscopic examination of faeces from domestic animals with diarrhoea. This led to the local recognition of coronaviruses in the faeces of scouring cattle[5] an of rotaviruses in calves with diarrhoea[2]. Since these initial reports, a large number of faeces from a variety of species of domestic animals have been examined, resulted in rotavirus being identified in calves, piglets, and foals[3], and coronavirus in cattle, lambs, deer and foals[3].

In an endeavour to obtain further information on the role of these two viruses in causing diarrhoea in calves, we recently examined a large number of faeces from known 'infected' properties, and from properties where the calf-diarrhoea status was unknown.

MATERIALS AND METHODS

Faeces Survey. Faeces samples were collected from 5 calves, 2-3 weeks old, from each of 30 properties in the practice area serviced by one of the authors (B.C.F.). The properties were selected without regard to disease status.

Known 'Infected' Properties. Three properties (E, F and G), where rotavirus infection had been demonstrated in the previous year, were selected for study, as was a further property (H) where coronavirus infection had been previously demonstrated. Faeces were collected from each of 10 calves at weekly intervals over a period of 5 weeks, commencing at 1-2 weeks old. All faeces were coded and processed 'blind'. Records were kept of clinical disease and treatments.

Virological Procedures. Faeces were made to a 10% suspension in cell culture medium, clarified by low-speed centrifugation, and the supernatant fluids centrifuged at 100 000 g for 1½ hours. The deposited pellet was resuspended in medium, stained with sodium phosphotungstate (pH 6.8) on a carvon-formvar grid, and examined under an electron microscope.

RESULTS

Faeces Survey. No viral particles, apart from bacteriophages, were recognised in 130 faeces from 26 of the properties. Rotavirus and coronavirus-like particles were found in calf faeces from four properties (Table I). When present, viral particles were found to be very numerous, except in the calf from property C, where very few rotaviruses were detected. The virus-containing faeces were of semi-fluid to watery consistency, and coloured grey to khaki.

Known 'Infected' Properties. Electron microscopic examination of calf faeces from those properties (E, F and G) where rotavirus infection had been previously demonstrated, showed that the onset of rotavirus excretion coincided with the onset of an outbreak of diarrhoea (*Fig. 1.*). Coronavirus-like particles were also detected in the faeces from two of these farms, but in only one did they coincide with the onset of diarrhoea.

Annual epidemics of calf diarrhoea had occurred on property E for 12 years. Rotavirus infection was first recog-

TABLE I: VIROLOGICAL FINDINGS ON 4 PROPERTIES FROM THE FAECES SURVEY

Property	Number of faeces positive: Rotavirus	Number of faeces positive: Coronavirus	Diarrhoea outbreak present
A	1	1	Yes
B	2	0	No
C	1	0	No
D	0	2	Yes

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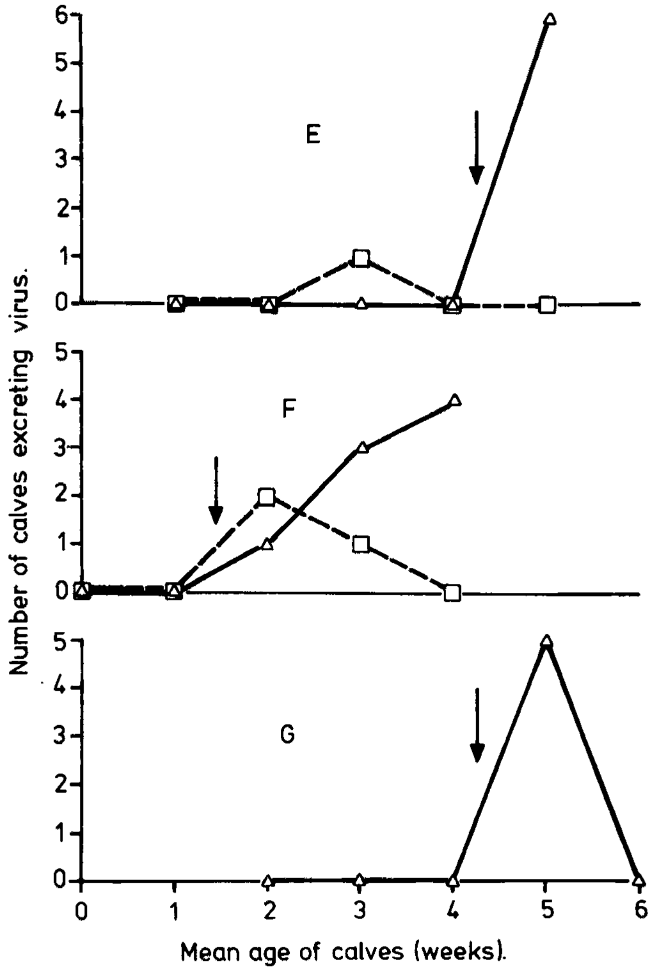

Fig. 1: Virological findings on rotavirus 'infected' properties over 5 week observation periods. (Arrow indicates the onset of outbreak of diarrhoea.

△———△ = rotavirus □———□ = coronavirus.)

nised in 1976, by electron-microscopic examination of calf faeces. Shortly after this diagnosis was made, a calf was sold to property G, where almost immediately afterwards, an 'explosive' outbreak of diarrhoea developed in the calves. Rotavirus was detected in a number of these calves, including the recently purchased animal. At both these farms, the outbreaks of diarrhoea have since continued to recur annually, with up to 80% of the calves being affected, despite good standards of hygiene and management. Affected calves became depressed, inappetant and rough-coated, and produced a white or green liquid faeces which was sometimes foul-smelling. Both hand-reared and suckled calves were affected, but more commonly the former. Colostrum, or colostrum-milk mixtures, were fed on both properties to calves for several days after birth.

On property F, where hygiene and management were poorer, diarrhoea outbreaks in calves were reported to have been a problem for more than 8 years. Almost all calves were affected as described above, but the disease appeared to spread more rapidly among younger calves, and resulted in a diarrhoea of a much more watery consistency. Up to 20% of calves died during the outbreak, and a further 20% were subsequently stunted in development. Colostrum was fed to calves during the first 48 hours only.

On property H, diarrhoea had been an annual problem in calves for several years, but only coronavirus-like particles had been previously demonstrated. During the period of observation, an outbreak of yellow water diarrhoea started when the calves were about 3 weeks old, and spread through the entire calf crop, resulting in 6% mortality but no stunting. Small numbers or coronavirus-like particles were detected at this time in the faeces of 1 of the 10 tested calves. On this property, calves were were taken from the cow 12 hours after birth, and were then bottle fed the dam's milk for up to a week. Though not part of the study, it was noted that, 2 months after the cessation of regular observation, a further outbreak of diarrhoea affected 12% of the calves on this farm. On this occasion, the submitted faeces samples were found to contain numerous rotavirus particles.

Treatment regimes instituted on the four properties varied somewhat, but generally consisted of oral antibiotics and/or sulphonamides, supplemented with oral glucose and water. Milk feeding recommended 1-2 days after clinical recovery. Though treatment did not prevent the onset and spread of the disease, it did reduce its duration to 2-3 days from the usual 1-3 weeks in untreated calves. On property H, treatment frequently eliminated diarrhoea in 24 hours.

DISCUSSION

Electron-microscopic examination of 150 faeces from 30 herds revealed that rotaviruses were present in 3 herds, and coronavirus-like particles in 2 herds. It is likely, however, that these levels err on the low side, due to the small number of calves examined from each property, the short duration of sample collection, the limited duration of virus excretion, and the limited sensitivity of the virological technique used. The results obtained, coupled with previously gathered data[2][3][5], indicate that both viruses may be widespread in the cattle population.

Based on overseas reports, rotavirus infection of cattle appears to be widespread in many countries[6], and can show a high level of prevalence. Woode[15] examined 59 cattle herds in the U.K., and found all of them to have serological evidence of rotavirus infection. Information on the distribution of coronavirus infections in cattle is more limited. However, the identification of coronavirus-like particles in cattle faeces in a number of countries[17] indicates that this virus is likely to be widespread also. Both viruses were concluded by Moon *et al.*[11] to be common amongst North American calves, based on published data.

In the known rotavirus 'infected' herds of this study, the onset of the outbreak of diarrhoea coincided with the onset of rotavirus excretion in the faeces, suggesting a cause-and-effect relationship. On the other hand, two of the three rotavirus-positive farms (B and C), detected during the faeces-survey, reported no problems with outbreaks of diarrhoea, though individual infected calves did show evidence of diarrhoea. The third rotavirus positive farm (A) detected during the survey did experience an outbreak of diarrhoea, but was complicated by a concurrent coronavirus infection. On property F, the onset of diarrhoea also coincided with the onset of excretion of a mixed infection of rotavirus and coronavirus-like particles, and was characterised by a much more severe clinical disease. However, as this property had poorer standards of hygiene and management, it is difficult to establish whether the more severe syndrome is a consequence of the double viral infection, or of other undiagnosed pathogens or management factors. A further complication is that the diarrhoea spread rapidly through the calves on this farm, yet the rotavirus infection appeared to build up more slowly.

Understanding the effect of the coronavirus-like agent is complicated by the contrary results obtained on two of the other farms. On property E, the virus was not associated with diarrhoea in the calves, in contrast to property F, where a severe outbreak of diarrhoea occurred coincidentally with the detection of the virus. The agent was, however, detected in only small numbers in one calf, and it is possible that a further undiagnosed agent was responsible.

Overall, the results indicated a clearer cause-effect relationship with rotavirus than with coronavirus. It is apparent, however, that a number of other factors may also be involved in the expression of clinical disease, to account for the variation in disease between properties.

The pathogenicity of rotaviruses and coronaviruses in calves has been well established[8][9]. It has been found, however, that clinical disease may not necessarily follow infection[6]. Manifestation of clinical disease by these agents has been found to be largely controlled by the amount of colostrum-derived antibody present in the intestinal lumen at the time of viral challenge, rather than by the level of systemic antibody[6][7]. In this regard, the earlier onset of diarrhoea seen in calves on property F may be due to the earlier cessation of colostrum feeding in comparison with properties E and G, so allowing earlier depletion of intestinal antibody levels and, hence, earlier susceptibility to acquired infection from the environment.

The protective effect of colostrum against rotavirus infection of the intestine has been found to be dependent on both the volume and the antibody titre of the ingested colostrum[6][11]. To be effective, it is necessary for colostrum to be fed very soon after calving, as the level of rotavirus antibody has been found to drop very rapidly within 48 hours of calving[4][14]. Low intestinal antibody levels may allow viral replication within the intestine which, if extensive enough, may result in clinical disease. It is likely also that when a level of immunity is present within the intestine, it may be overcome by the effect of a heavy viral challenge. The levels of rotavirus excreted in infected faeces may be very high and, as the virus is comparatively stable, they may result in a very heavy contamination of the environment[16]. On properties E and G, hand-reared calves were apparently more commonly affected than naturally suckled calves. This may be a result of a higher build-up of virus in the environment of the hand-reared calves due to more intensive stocking, in comparison with calves run at pasture with their dams.

Though it has been suggested that various strains of rotavirus possessing different levels of virulence may account for the variable expression of the disease, direct evidence for the existence of such strains has as yet not been presented[6].

It has often been suggested that mixed infections may result in enhanced pathogenic effect. There is, however, little evidence to support or deny this view from the published data[11]. Nevertheless, Waldham *et al.*[13] reported that combined infections of rotavirus and *Providencia stuartii* resulted in enhanced pathogenicity. Whether or not this occurs with other combinations of organisms can only be established by further studies.

ACKNOWLEDGEMENTS

We thank Miss C. Watters for her excellent technical assistance and also the numerous farmers who co-operated in the project.

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